stain), iliac neointimal hyperplasia (I/M ratios) and Mac density (RAM-11 stain) were examined. Macs decreased significantly in groups treated with the combination of MGd and Asc, more than with Asc alone (Table). *In vitro*, cellular uptake of MGd (50 µM/24h) by flow cytometry (fluorescence>650nm) showed median fluorescence values of 33.47±0.93, 26.46±1.40 and 6.48±0.97 (mean±SD) in human umbilical vein endothelial, coronary artery endothelial and smooth muscle cells respectively and 23.88±0.08 in Macs (THP-1 cells). **Conclusions**: MGd uptake occurs in human vascular cells and Macs. Activation of MGd by Asc decreases Macs in rabbits, probably as a consequence of futile redox cycling and ROS generation. This approach offers promise for the treatment of vulnerable plaque.

Table. Study Results (mean±SEM)

Groups	Control	MGd	Asc	MGd+Asc(-4h)	MGd+Asc(0 h)	MGd+Asc(+4 h)
Aortic plaque(%)	33.8±8.8	37.9±11.9	39.6±9.7	32.8±6.1	38.2±6.1	23.4±6.4
Iliac I/M ratios	2.2±0.1	2.4±0.2	2.7±0.2	2.3±0.2	2.1±0.2	2.6±0.2
Iliac Macs(%)#	30.3±1.3	31.7±1.2	**21.4–2.1***	**13.0–1.1***	**12.6–1.1***	**15.7–1.2***

*# % total intimal area, p<0.001 by oneway ANOVA; * p<0.05 compared to control*

POSTER SESSION

1103 Vascular Structure and Function

Monday, March 31, 2003, Noon-2:00 p.m.
McCormick Place, Hall A
Presentation Hour: 1:00 p.m.-2:00 p.m.

1103-126 Endogenous Estrogens Influence Endothelial Function in Young Men

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Background: Estrogen is known to influence cardiovascular function in women. Males produce endogenous estrogen from testosterone via the enzyme aromatase. Previous studies have suggested a role for endogenous estrogens in cardiovascular function in men. We therefore examined the effects of reducing endogenous estrogen production by aromatase inhibition on vascular function and lipid levels in healthy young men.
Methods: A double blind placebo controlled randomized design was used. Twenty healthy men, aged 18 to 32, were randomized to receive either the aromatase inhibitor, anastrozole (1mg) or matching placebo. Sex hormone and lipid levels were measured. Endothelial function was assessed by flow mediated dilation (FMD) of the brachial artery in response to ischemic hyperemia of the forearm, and endothelium independent vasodilation was assessed as the effect of sublingual glyceryl trinitrate (GTN) on brachial artery diameter. Measurements were made at baseline and after 6 weeks of treatment.
Results: There was one withdrawal in the treatment group, for unspecified reasons. Compared to baseline, a significant decrease in FMD was observed in subjects on anastrozole, median 6.1 (5.2-13.4) to 3.5 (2.0-5.7) (p=0.034), but there was no difference in the placebo group. No changes were observed in GTN-induced endothelial independent dilation following either anastrozole or placebo. There was a significant decrease in estrogen concentrations with aromatase inhibition, mean 85.4 ± 4.2pmol/L to 64.3 ± 8.1pmol/L (p=0.042). There were no significant changes in cholesterol, triglyceride, testosterone or DHEA levels in either the anastrozole or placebo group.
Conclusion: Selective suppression of endogenous estrogens by inhibition of aromatase in healthy young men results in impairment of flow mediated vasodilation, a response known to be mediated via local nitric oxide release. Our results suggest a role for endogenous estrogens in endothelial function and vascular health in young males.

1103-127 Role of Cytomegalovirus and Asymmetric Dimethylarginine in the Derangement of Nitric Oxide Synthase in Cardiac Allograft Recipients

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Background We hypothesized that cytomegalovirus (CMV) induced increases in ADMA (asymmetric dimethylarginine, the endogenous inhibitor of NO synthesis) may contribute to the coronary endothelial dysfunction observed in cardiac allograft recipients.
Methods and Results: Forty orthotopic heart transplant recipients without acute rejection or infection episodes were studied. Heart transplant recipients manifested elevated ADMA levels (by HPLC) compared to non-transplant controls. Transplant patients with CMV DNA positive leukocytes (by PCR) had higher ADMA plasma concentrations and more extensive coronary artery disease than transplant patients without leukocyte-associated CMV DNA. Four weeks after heart transplantation, five recipients , (male, mean age: 50.6 + 6.2 years) underwent coronary catheterization. Acetylcholine (ACH) and nitroglycerin (NTG) were infused into the LAD , and quantitative angiography was performed to assess vascular reactivity immediately before and after Ach Infusion, aortic and coronary sinus blood was drawn for calculation of basal and stimulated plasma NOx with Griess Assay. ACH caused paradoxical vasoconstriction (3.32 + 0.09 mm vs 2.83 + 0.29 mm), whereas NTG induced significant dilation (2.61 + 0.17 mm vs 3.61 + 0.18 mm, P < 0.01).The plasma NOx levels in the AIV were not increased by ACH infusion in LAD (40.4 + 7.8 Um vs 39.1 + 7.9 uM) indicating a severe impairment in NO synthesis.
Conclusions: Synthesis of endothelial nitric oxide is severely depressed in the coronary arteries of heart transplant patients. This abnormality may be secondary to high plasma levels of the circulating NOS inhibitor, ADMA. CMV infection exacerbates this abnormality.

1103-128 Coronary Endothelial Foot Print of Vasa Vasorum Perfusion Territories: Micro-Computed Tomography Analysis of Porcine Coronary Arteries With and Without Microembolization

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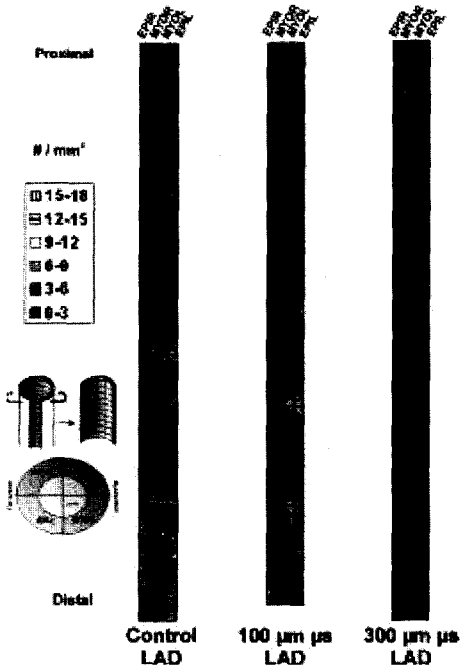
Background As removal or ligation of vasa vasorum (VV) has been shown to cause atherosclerotic lesions, it seems likely that microembolization (ME) of coronary artery VV should also. This study starts to examine the question as to whether individual VV perfusion defects in the coronary artery wall due to ME occur in plaque-like distributions.
Methods Non-radioactive microspheres (µs) 300 or 100µm in diameter were injected in 3 porcine LADs each. After harvesting the heart, the LADs were injected with radiopaque Microfil®. Up to 10 cm-long segments of the LADs were removed and scanned intact with micro-CT (20 µm cubic voxels). The spatial density (#/mm²) of VV was measured in 20 µm-thick cross-sections (spaced at 0.8 mm intervals, distal to the µs-injection-site and in corresponding segments of 3 control LADs). These cross-sections were subdivided into 2 epicardial (epi) and 2 myocardial (myo) quadrants.
Results ME reduced VV densities (3.75±2.03 in controls vs 3.06±1.26 in 100 and 2.50±1.44 in 300 µm µs; all P<0.001) and resulted in patchy distribution of longitudinal and circumferential VV densities (Fig.). Epi VV densities were consistently higher than myo densities (4.49±3.27 vs 2.88±2.56 in controls; 3.57±2.22 vs 2.45±2.10 in 100 and 3.21±2.76 vs 1.75±2.16 in 300 µm µs; all P<0.001).
Conclusion Microembolization could be a proatherogenic factor by focally impairing coronary artery wall perfusion. Our data are also consistent with preferential plaque formation at the myocardial side of coronary arteries.



1103-129 Coronary Endothelial Dysfunction Is Associated With an Increased Risk of Cerebrovascular Events

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Background: Stroke, due to atherothrombotic disease, represents a leading cause of disability and death in the western world. Endothelial dysfunction, which is considered a key factor in atherogenesis, is associated with an increased risk of cardiovascular events. However, the magnitude of the association between coronary endothelial dysfunction (CED) and cerebrovascular events (CVE) is unknown. This study was performed to investigate the association between CED and CVE.
Methods: We studied 503 consecutive patients without obstructive coronary artery disease (CAD) who underwent coronary endothelial function testing. Patients were stratified according to the presence (n=305) or absence (n=198) of CED. Medical records were examined for the occurrence of CVE defined as ischemic or hemorrhagic stroke or transient ischemic attack either preceding or following coronary endothelial function testing.
Results: A total of 25 cerebrovascular events were documented, 22 in patients with CED and 3 in patients without CED (p=0.008). Multivariable logistic regression, which included demographic characteristics and traditional cerebrovascular disease-related risk factors, identified the presence of CED as the single strongest factor associated with CVE (odds ratio: 4.32; 95% CI, 1.26-14.83). Kaplan-Meier analysis demonstrated a significantly

worse CVE-free survival for patients with CED (p=0.04).
Conclusion: Presence of CED in patients without obstructive CAD is associated with an increased risk of CVE. Detection of this early stage of atherosclerosis may provide important information to identify patients who would benefit from aggressive preventive strategies.

1103-130 Postprandial Endothelial Dysfunction Is Not Apparent in Young Healthy Individuals

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Background : The intake of a fatty meal acutely impairs endothelial function and this mechanism may partially explain the atherogenic role of postprandial lipemia (PL). Since age is another important determinant of endothelial function, we assessed postprandial endothelial dysfunction in two age groups of healthy volunteers.
Methods : We measured serum lipoproteins and brachial artery flow-mediated dilation (FMD) (an index of endothelial dysfunction) in 2 groups of healthy individuals before and 2 and 4 hours after one 50 gr saturated fat meal. Group A consisted of 14 middle-aged volunteers (45.14 ±6.21 years old, 11 men and 3 women) and group B consisted of 14 young persons (25.71±5.4 years old, 11 men and 3 women). Brachial artery FMD was assessed with the use of a 7.5MHz vascular ultrasound transducer. Statistical analysis was done with Friedman two-way analysis of variance.
Results : Lipid profile, baseline brachial artery diameter and baseline FMD were similar in both groups. In both groups, the fatty meal increased triglycerides (119.2±66.3 to 160.2±80.2 to 179.9±105.5 mg/dl, p=0.0006 for group A and 82.8±37 to 118±45.5 to 138.6±56.3 mg/dl, p= 0.0001 in group B). LDL-cholesterol was significantly decreased only in group B (127.4±45.4 to 120.4±42 to 118.4±37.4mg/dl, p=0.03 in group B vs 134.4±36.3 to 125.7±35 to 124.9±36.7mg/dl, p=0.2 in group A) .The rest of the lipoproteins did not change postprandially in either group. Brachial artery FMD was significantly reduced only in group A individuals (15±8% to 11±7% to 10±3%, p=0.012) while in group B it remained relatively unchanged (15±4% to 13±4% to14±7%, p=0.6).
Conclusions: Our findings further support the hypothesis that a meal with high content in saturated fat acutely impairs endothelial function of peripheral blood vessels in healthy subjects. However, this effect is not apparent in young persons. Other regulatory mechanisms, possibly associated with a more favorable postprandial lipid profile in the young persons, may account for this phenomenon.

1103-131 Are Endothelial Dysfunction and Inflammation Independently Related to Sleep Apnea Severity?

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Background Obstructive sleep apnea (OSA) is associated with cardiovascular disease (CVD), but the nature of this association is incompletely understood. Endothelial dysfunction and inflammation are recently recognized risk factors for the development of CVD. We tested the hypothesis that indicies of endothelial dysfunction (flow mediated vasodilation, peak hyperemic flow) and systemic inflammation (high-sensitivity C-reactive protein, hs-CRP) are increased in proportion to OSA severity.
Methods 130 subjects from the Cleveland Family Study (OSA patients and family members, community controls) were prospectively studied with overnight polysomnography, brachial artery ultrasonography (10 MHz, Acuson Aspen™), and hs-CRP in a clinical research facility. OSA was characterized by the apnea/hypopnea index (AHI). Outcome measures included percent changes in flow mediated dilation (ΔFMD) and peak hyperemic flow (ΔPBF), and log-transformed hs-CRP. Relationships between the AHI and outcome measures were assessed with univariate and multivariate analyses adjusting for age, race, sex, and obesity.
Results The study population was diverse (51% African American, 51% female), young (48 ±18 yrs), and obese (body mass index 34 ±11 kg/m^2). Univariate analyses showed that increased AHI was associated with lower levels of ΔPBF (r=-0.46) and ΔFMD (r=-0.30), and with higher levels of hs-CRP (r=0.34) (all p's <0.005). After multivariate analysis a significant negative relationship persisted between AHI and ΔPBF (p<0.05). The relationship between AHI and hs-CRP, while significant after adjustment for age, race, and sex (p<0.005), was attenuated after adjustment for obesity (p=0.20). Excluding subjects taking medications from the analysis and considering hypertension and diabetes as covariates did not materially alter the results.
Conclusions Hyperemic brachial artery flow, but not flow-mediated vasodilation, is reduced in OSA in a dose-dependent fashion. Elevated hs-CRP levels occur in OSA but this is partly explained by obesity. These findings suggest that systemic inflammation and resistance vessel endothelial dysfunction may contribute to OSA-related CVD.

1103-132 Pulse Wave Intensity: A New Parameter for Understanding Dynamic Ventriculoarterial Interaction

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Wave intensity (WI) is calculated as the product of the derivatives of velocity and pressure (or diameter
in non-invasive studies). It measures the instantaneous balance between forward and backward waves
travelling from the heart and reflected from the periphery. To determine if WI can provide a new clinical tool for assessing ventriculo-arterial interaction,a central dynamic process in hypertension and heart failure, we determined the feasibility of recording WI and measured its intra- and inter-observer and temporal variability.
Methods: The right common carotid artery was imaged (7.5MHz linear array probe). Diameter (from wall tracking of anterior and posterior wall displacements) and flow (from integrals of colour flow Doppler) were measured at the same site (Aloka SSD-5500). WI was calculated off-line; variability is reported as coefficients of variation.
Feasibility is reported from 115 subjects and variability from 61 normal subjects (median age 34.0yr) who were each studied by 2 trained observers on 2 occasions 2 weeks apart.
Results: WI could be measured in 96% of subjects. The first peak of WI coincides with acceleration and increasing pressure during early systole, and is a forward compression wave reflecting LV contraction; CV for intra-observer reproducibility were 5.3 and 2.2%. Inter-observer and temporal variability were greater (CV 12% and 31%). A second peak of WI is caused by a forward expansion wave related to deceleration in late systole; its intra-observer CV were 24.1 and 21.9%. A negative area in mid-systole is determined by wave reflections from the periphery and influenced by vascular resistance; intra-observer CV were 33.1 and 34.0%. An index of arterial stiffness (beta) derived from WI had intra-observer CV of 19.3 and 13.7%; inter-observer and temporal CV were 13.2% and 25.4%.
Conclusion: Non-invasive assessment of WI is feasible and gives reproducible information about wave travel in early systole and about arterial stiffness. WI in late systole is affected by considerable biologic variation. WI may give new insights into changing ventriculo-arterial interaction during treatment of hypertension and heart failure.

1103-133 Reactive Oxygen Species Are Involved in Smoking-Induced Dysfunction of Nitric Oxide Biosynthesis and Upregulation of Endothelial Nitric Oxide Synthase in Human Coronary Artery Endothelial Cells

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Background Our group has previously demonstrated that smokers' serum incubated with human umbilical vein endothelial cells (HUVECs) reduced nitric oxide (NO) availability and endothelial nitric oxide synthase (eNOS) activity in the presence of increased eNOS expression. Whether these observations extend to human coronary artery endothelial cells (HCAECs) is unknown. Additionally, the potential role of reactive oxygen species is also unclear.
Methods and Results Confluent (~85%) monolayers of HCAECs were incubated with serum from 9 nonsmokers and 15 smokers for 12 hours with or without the addition of free radical scavengers, cell-permeable polyethylene glycol-superoxide dismutase (PEG-SOD, 300U/mL) or PEG-SOD+PEG-catalase (1000U/mL) or tetrahydrobiopterin (BH_4, 20uM)(an essential co-factor for eNOS) treatment. At the end of incubation, NO availability, eNOS protein, and eNOS activity were measured from the same culture by standard techniques. HCAECs incubated with smokers serum alone showed significantly lower NO level (0.02±0.01 versus 0.07±0.01 uM/pg eNOS/mg total protein, P<0.007), higher eNOS expression (3908±269 versus 2182±281 pg eNOS/mg total protein, P<0.005) but lower eNOS activity (0.25±0.03 versus 0.50±0.08 pmol L-citrulline/min/pg eNOS/mg total protein, P<0.005) compared to nonsmokers similar to our previous findings in HUVECs. In smokers, PEG-SOD or PEG-SOD+PEG-catalase or BH_4, all significantly (P<0.05) improved NO availability and eNOS activity. A significant decrease in eNOS expression was only seen with PEG SOD+PEG-catalase treatment (P<0.005) while PEG-SOD alone trended to increase eNOS expression. In nonsmokers all of the above treatments had no significant effect on any of the parameters.
Conclusions These data in HCAECs confirm our previous findings in HUVECs and suggest that oxidative stress plays a central role in smoking-mediated dysfunction of NO biosynthesis. Various free radical scavengers may act differently to improve dysfunctional NO biosynthesis.

1103-134 Impaired Endothelium-Dependent Vasomotion in Patients With Recent Myocardial Infarction and Hyperhomocysteinemia

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Backgound: Hyperhomocysteinemia (HHC) is a pro-thrombotic condition that may cause oxidative endothelial injury and impaired endogenous fibrinolysis. We tested the hypotheses that (1) patients with recent myocardial infarction (MI) and HHC demonstrate impaired endothelium-dependent vasomotion and endogenous fibrinolysis, and (2) vitamin supplementation reverses endothelial dysfunction in HHC. **Methods:** Plasma homocysteine (HC) was determined in 120 patients admitted with MI. From the upper and lower plasma HC quartiles, 18 patients were recruited into a randomized double-blind placebo-controlled crossover trial at least 4 months after the index event. Patients were studied on 2 occasions after a 4-week course of placebo or folate (5 mg)/cyanocobalamin (100 μg)/pyridoxine (10 mg) tablets. Bilateral forearm blood flow (FBF) was measured using venous occlusion plethysmography during intra-arterial infusion of substance P (4-16 pmol/min), acetylcholine (5-20 μg/min) and sodium nitroprusside (2-8 μg/min). Venous samples were assayed for tissue plasminogen activator (t-PA) antigen and activity. **Results:** Patients in the upper HC quartile had higher plasma HC concentrations (16.8±2.9 vs 7.9±0.7 μmol/L; P=0.003). Vitamin treatment resulted in an increase in serum vitamin B12 and greater than 2-fold increase in serum folate (P<0.05) but did not reduce HC concentrations. All vasodilators caused dose-dependent increases of FBF in the infused arm (P<0.05). FBF response to acetylcholine but not sodium nitroprusside was reduced in HHC patients compared to control patients (5.1±1.2 vs 8.0±1.5 ml/100ml/min; P=0.01). There was no difference in substance P-induced t-PA release in HHC patients and vitamin treatment did not affect FBF responses or t-PA release. **Conclusion:** We conclude that HHC is associated with impaired endothelium-dependent vasodilatation but no alteration in acute endogenous fibrinolysis in patients with recent MI.